Microscopy without imaging: compressive sensing for heart-synchronized imaging

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Abstract: We demonstrate experimentally that direct analysis of compressively sensed signals provides sufficient information to achieve high-precision phase lock to a periodically-moving structure, without any need to ever reconstruct an image of the target object.

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1. Introduction

Video images consist of tens of thousands of pixels, but often the useful information we wish to obtain from image analysis is much simpler. For example, a target recognition problem may require an object to be classified into one of a small number of categories [1], we may require an estimate of local motion in a scene [2], or we may wish to phase-lock to a periodically-moving structure such as the heart [3]. This latter scenario is the focus of the present manuscript: we will show how complex information can be calculated directly from a small number of compressive single-pixel measurements.

A traditional image analysis pipeline would start with image formation, followed by some form of image analysis to extract the desired information. These images would normally be formed by measuring directly in the pixel basis (e.g. on a CCD), but the frameworks of computational imaging and compressive sensing offer a different approach to image formation. Here individual scalar measurements can be made in a different basis, often a global basis where every measurement captures partial information about every pixel in the object. This gives the potential for imaging in a wider range of spectral bands [4] not limited by the availability of large pixel array imagers, frees us from the constraints of a regular pixel grid [5], and opens up the possibility of *compressive* imaging and reconstruction. Here, known information or assumptions about the characteristics of the scene can be incorporated into the reconstruction as *priors*, making it possible to reconstruct an image from a set of sub-Nyquist sampled measurements.

However, in the visible waveband there is often little to be gained from the additional complexity of a compressive *imaging* system, where even a factor of 10-100 reduction in the number of sequential, compressive measurements would not offer serious competition to the simplicity of a low-cost machine vision camera. This assumes, however, that it is *necessary* to reconstruct a high-quality image before image analysis can commence. If the analysis stage can be performed through direct processing of the compressive measurements, the computationally-demanding reconstruction can be bypassed entirely. We will demonstrate practical application of this concept to the problem of optical heartbeat-synchronization for *in vivo* zebrafish microscopy. In this scenario, the conventional approach involves acquiring microscope images in transmission mode, which are then are analyzed to determine the "phase" of the heartbeat as a function of time. This information is used to generate electronic signals to trigger acquisition of fluorescence images at the correct point in the heartbeat [3]. However, instead we will directly analyze 7 single-pixel measurement channels, a sufficiently low number that it would be impossible to reconstruct anything remote resembling an image of the scene, and yet we will show that we can derive accurate phase information from these massively sub-sampled measurements.

2. Optical and computational design

It has previously been shown that a linear function of some input image can be efficiently estimated using a small number of compressive measurements [6]. In our case our desired quantity is the phase of the heart, which is a highly nonlinear function of the input data. We wish to make a small number of measurements of our object in a suitable basis, and process these using a target recognition algorithm that can determine the heartbeat phase as a function of time [3]. Our algorithms can then make forward predictions of when the heart will be at the correct point in its cycle, and trigger acquisition of a heartbeat-stabilized fluorescence image.

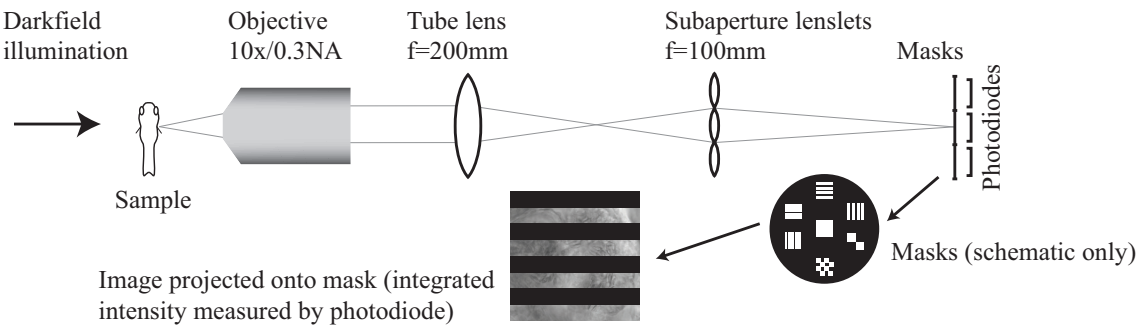


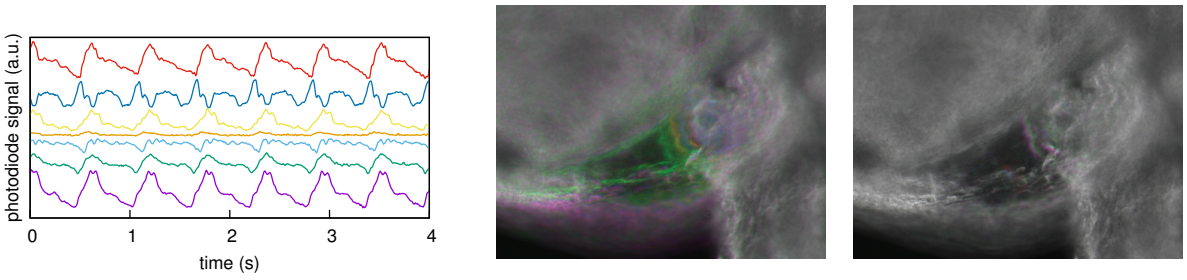
Fig. 1. Schematic optical setup, showing lenslet array projecting subaperture images onto chrome-on-glass mask, with transmitted intensity measured using an array of seven photodiodes. Illustrative chrome-on-glass mask shown, but for experimental data shown later, binary grating masks were used with a range of spatial frequencies.

Our optical design is based on 7-way image replication by pupil division. Each sub-aperture image is projected onto a different chrome-on-glass binary intensity mask, and the light transmitted through the mask is measured on a photodiode (Fig 1). Each photodiode makes a single scalar measurement in our compressive measurement basis. For the binary masks that define this basis, we selected highly regular grating and checkerboard type patterns, consisting of a variety of spatial frequencies. This meant that even though the signals would change if the object was in a different location in the scene, performance could reasonably be expected to be independent of object position. This then avoids the need for positioning the object precisely in the field of view, and also reduces the risk that computer-based modelling and optimization of our choice of measurement basis would be biased by the specific test imagery we used.

Preliminary computer modelling confirmed that our existing heartbeat-phase analysis algorithms [3] would in principle be capable of phase recovery from as few as 5 photodiode-based measurement channels. Practical challenges we encountered included electromagnetic interference, and the low SNR issues that are common in the types of global scene measurements commonly performed in compressive imaging. This is compounded by the fact that intensity-based measurements suffer from an inability to measure basis functions containing negative values, which in practice means that single raw measurements represent the linear sum of the basis function of interest (e.g. a sine function) and a constant offset (the zero-spatial-frequency component of the object). We overcame these issues by operating the microscope in a darkfield imaging mode, electromagnetic shielding of our readout circuitry, and signal filtering to eliminate residual 50 Hz electromagnetic interference. With these measures in place, we have been able to show that we can perform heartbeat-phase recovery and forward prediction with our 7 compressive measurement channels, in spite of this residual noise and natural random variations of the specimen being imaged.

3. Results

Fig 2(a) illustrates the photodiode signals measured for our seven compressive channels and sampled at 200 Hz (with 50 Hz noise filtered out as noted above). For validation purposes, this was accompanied by simultaneously-recorded widefield video images at 80 fps. Although no truly definitive phase reference is available for the heart, we considered a postacquisition analysis [7] of this widefield video as a reference standard to which our analysis of the compressive channels could be compared. First we compared a postacquisition analysis of the compressive channels to the equivalent analysis of the full widefield video to confirm that the information available in the seven photodiode channels is sufficient to recover accurate phase information. In this type of postacquisition analysis, the phase progression is assumed to be piecewise linear, and thus a measure of accuracy is the magnitude of the discontinuity between successive sections. Our initial 7-channel results had a RMS phase discontinuity of 0.04 (0.6% of a heartbeat), barely any worse than the value of 0.03 (0.5% of a heartbeat) from analyzing the reference widefield images. Next we tested our existing realtime synchronization algorithm, which performs forward prediction from the data received. We can estimate the triggering accuracy by examining the phases (obtained from the widefield validation channel) for the trigger times generated by our realtime synchronization algorithm. By this measure, the standard deviation of the phase at the calculated trigger times is only 0.12 (2% of a heartbeat), representing effective synchronization. Fig 2(b),(c) show the consistency of the synchronized images between heartbeats.



(a) (b) (c)

Fig. 2. (a) photodiode signals showing diversity of information available from different spatial frequency masks, ranging from no mask (bottom), 12.5 lines/mm (next up) to 200 lines/mm (top). (b) Three-colour composite RGB image superimposing three different phases in the heartbeat to illustrate the level of motion involved during the heartbeat. (c) Equivalent composite RGB image of three images (taken for validation) at the times calculated by our compressive phase-locking system. The uniform grey colour indicates that the images are effectively identical, all being taken at the same point in the heartbeat – this confirms the quality of the synchronization.

4. Conclusion

We have shown that 7 single-pixel measurement channels are sufficient to perform realtime phase analysis of a periodically-moving structure (the beating zebrafish heart), suitable for use in for realtime synchronized imaging. Since our method uses direct compressive measurement without image reconstruction, it has very low computational demands – the computation load is reduced by a factor of 10^4 compared to conventional imaging. The data processing can therefore be implemented on a low-cost microcontroller device, and the minimal data bandwidth makes our approach suitable for very high-speed and high-throughput image processing and measurement scenarios.

We found that, perhaps surprisingly, the same phase locking code we previously developed for processing full images is effective when applied directly to the compressively-measured signals. Although our algorithms could undoubtedly be specifically optimized for the compressive scenario, it is encouraging to find that existing algorithms perform effectively without any modification. Our work illustrates that direct compressive measurement of complex nonlinear quantities derived from a scene can be performed with much reduced computational cost – a result with broad implications for object recognition and tracking, motion detection, and image analysis.

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